

4th Generation Testing: What Does It All Mean?

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We'll Be Discussing...

- 4th Generation Conventional Testing
 - What it is
 - How to interpret results
 - How to counsel clients regarding results

Testing Has Evolved...

- That's a good thing!
- What's still the same?
 - Conventional blood draw and rapid test options still exist...

What's Changed?

- 4th generation testing identifies infection sooner than earlier tests
- Multispot is no longer in production
- Geenius is an automated test that produces additional results not previously available with Multispot
- We now have a broader algorithm to guide medical management decisions

How Is Infection Identified?

- IgM antibodies: produced soon after exposure to an infection
 - Usually present about 3 weeks after infection
- IgG antibodies: a later response to infection; a long term response to infection
 - Usually present about 6 weeks after infection

What Is p24 Antigen?

- Viral protein that makes up most of HIV-1 viral core
- Serum concentrations of p24 antigen high in first few weeks after infection & appear again in end stage HIV infection
- Tests sensitive to p24 antigen useful for diagnosing very early infection (i.e., acute) when antibody levels are below antibody-based assay detection limits

How Is Acute Infection Identified?

- Identification of HIV RNA in the absence of antibodies
- HIV-1 RNA is first detectable approximately 10 days after an individual becomes infected
- Detectable by HIV-RNA Nucleic Acid Amplification (NAT) test

HIV Testing Through the “Generations”

Screening Tests

EIA (Enzyme Immunoassay)

- **1st Generation EIA (1985)**
 - Detects HIV-1 IgG Antibody
- **2nd Generation EIA (1991)**
 - Detects HIV-1 IgG Antibody
- **3rd Generation EIA (1992)**
 - Detects HIV-1/HIV-2 IgM & IgG Antibodies
- **4th Generation EIA (2010)**
 - Detects HIV-1/HIV-2 IgM, IgG Antibodies **PLUS** p24 Antigen, however does NOT distinguish which test component of the assay is positive

Old Algorithm

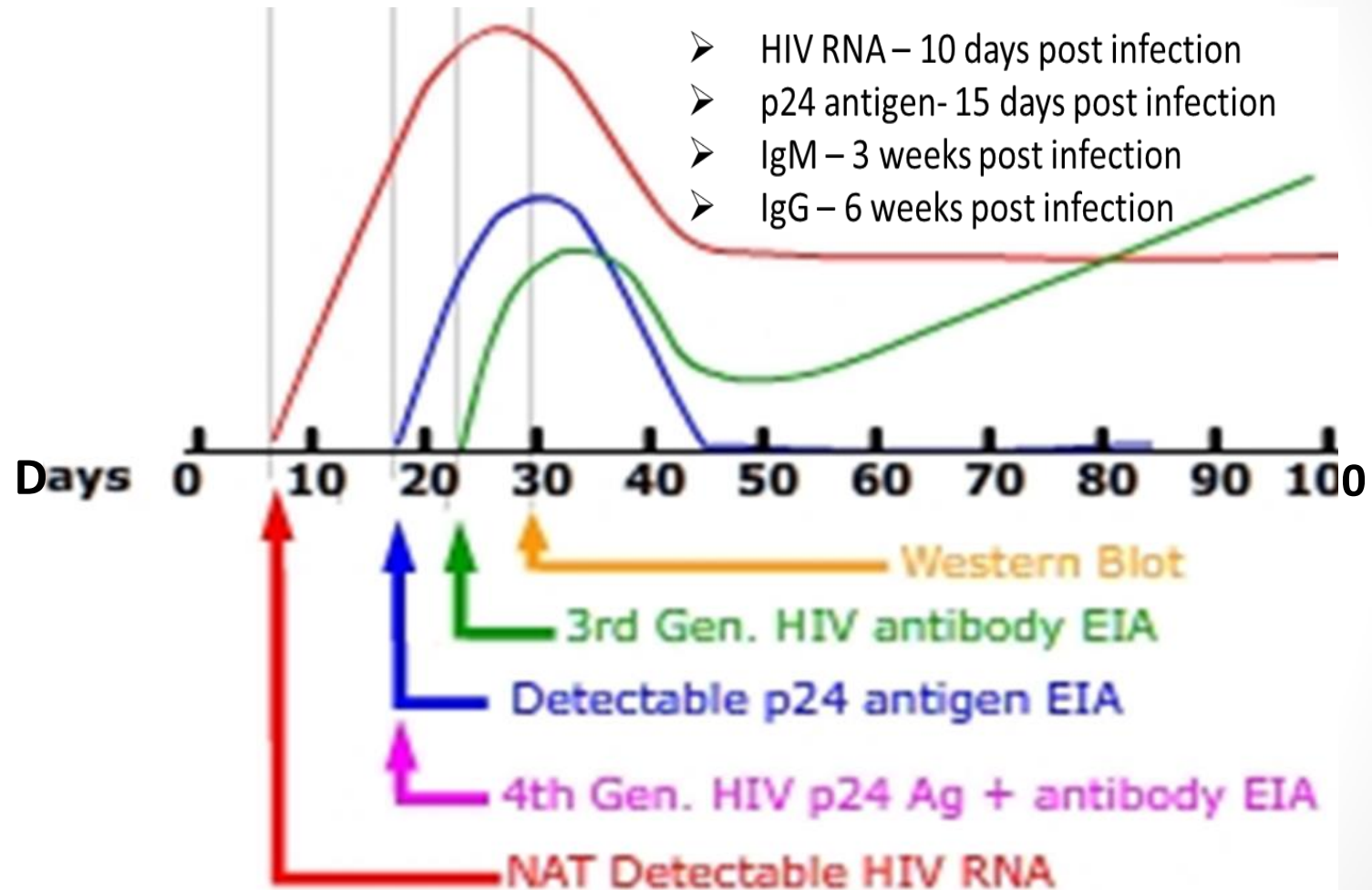
**Confirmation
of HIV-1 by
Western Blot
Assay**

New Algorithm

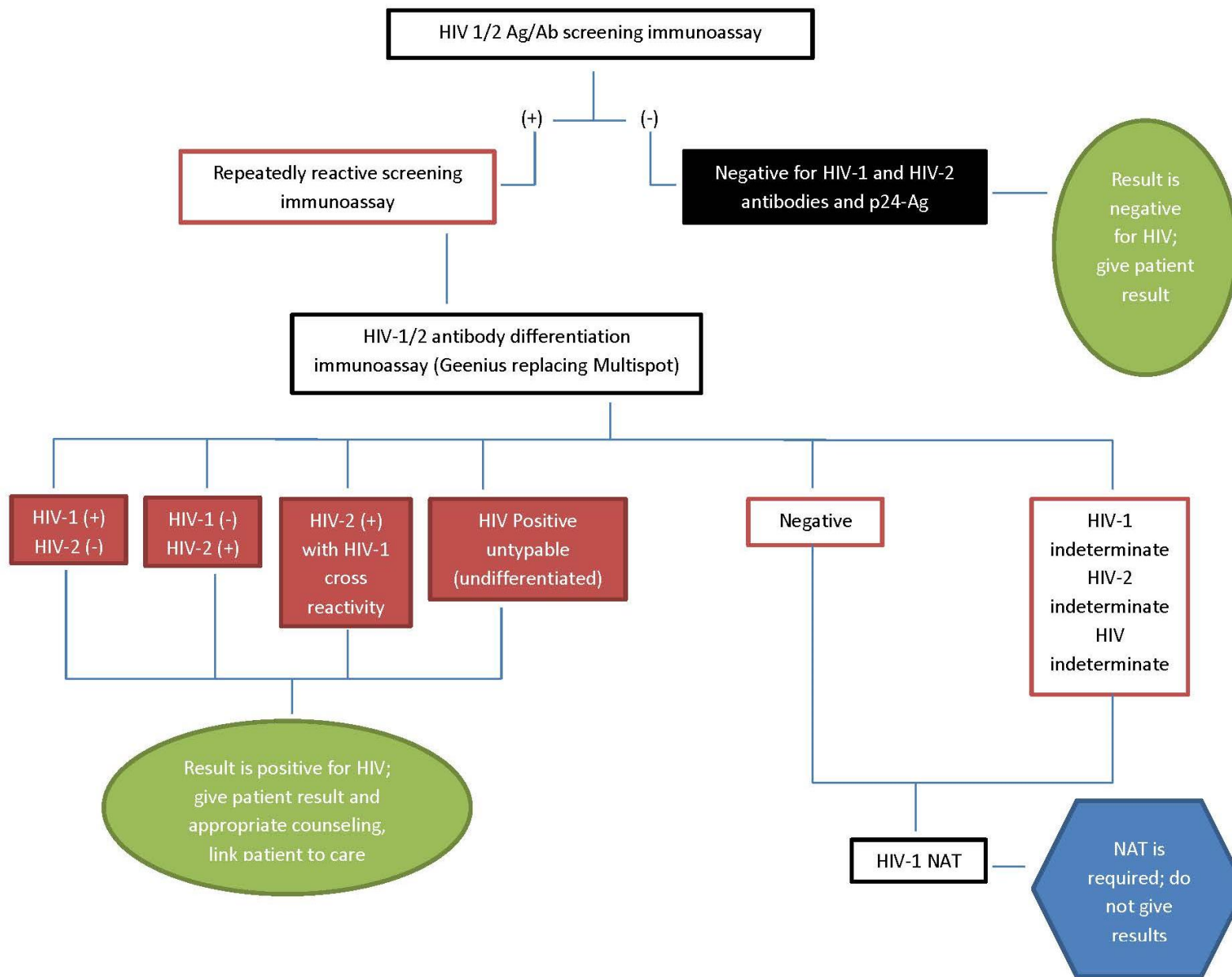
**Identification of
HIV-1 & HIV-2 by
Differentiation Assay
(Geenius)**

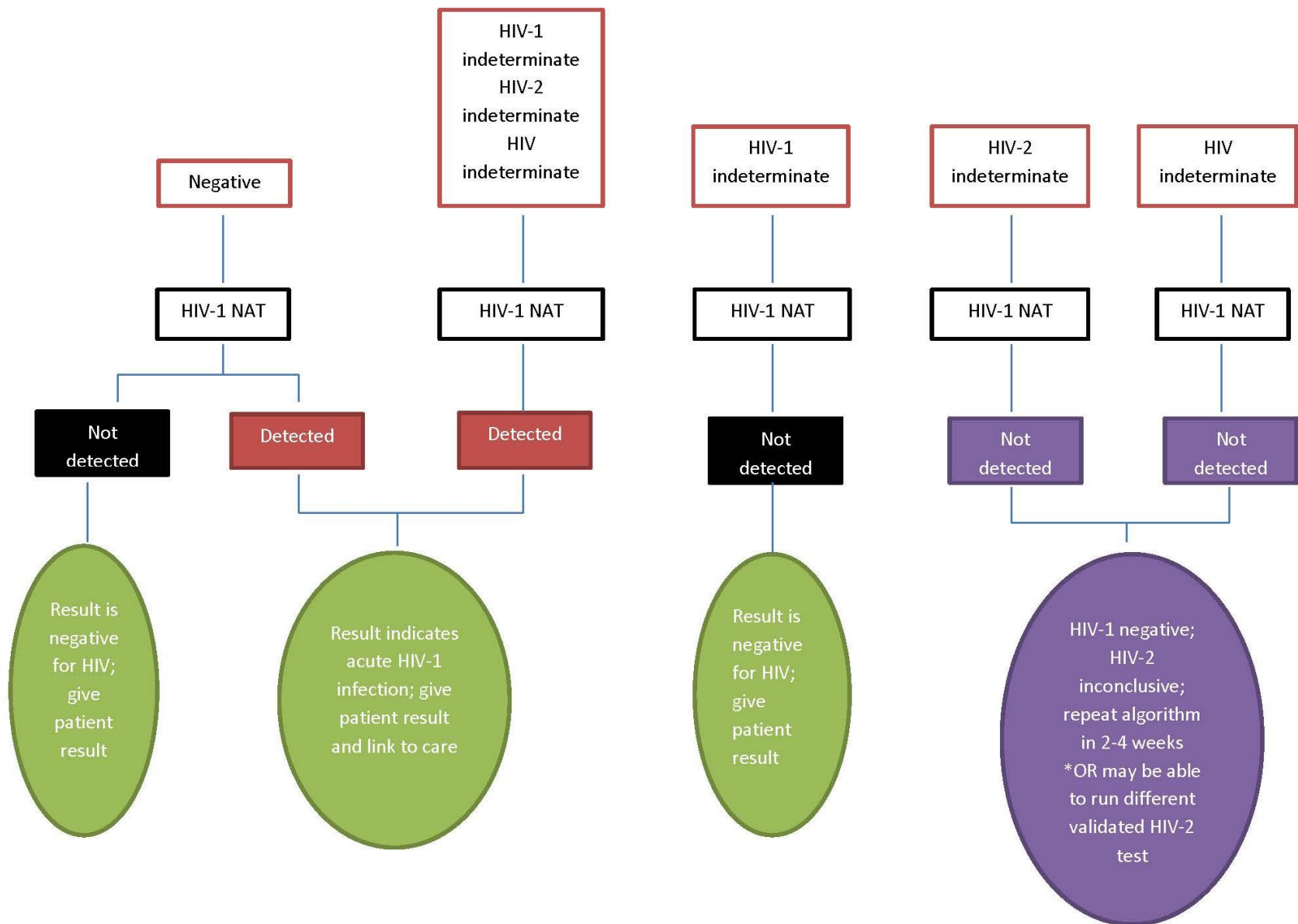
“Window Period”

Infection to Detection

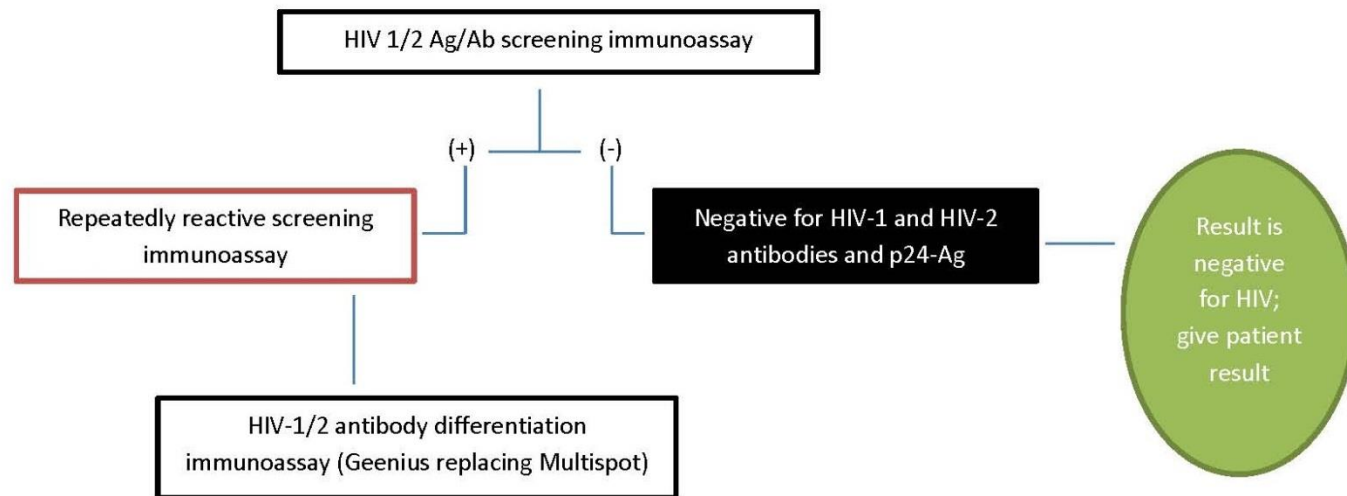


“Window Period” is the time frame between initial exposure and the time it can be reliably detected. Window Period aka Eclipse Phase.

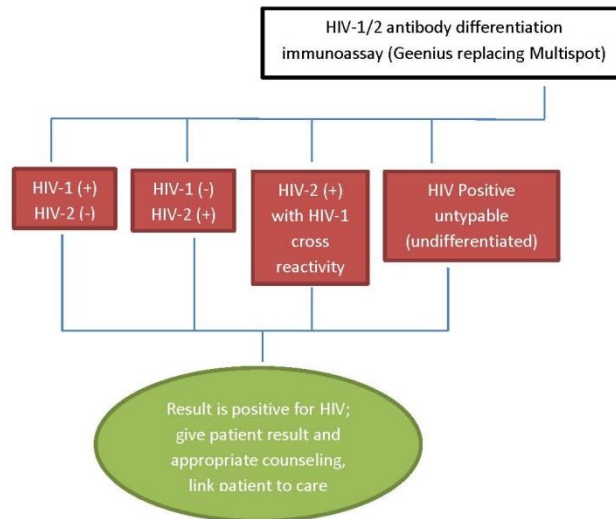




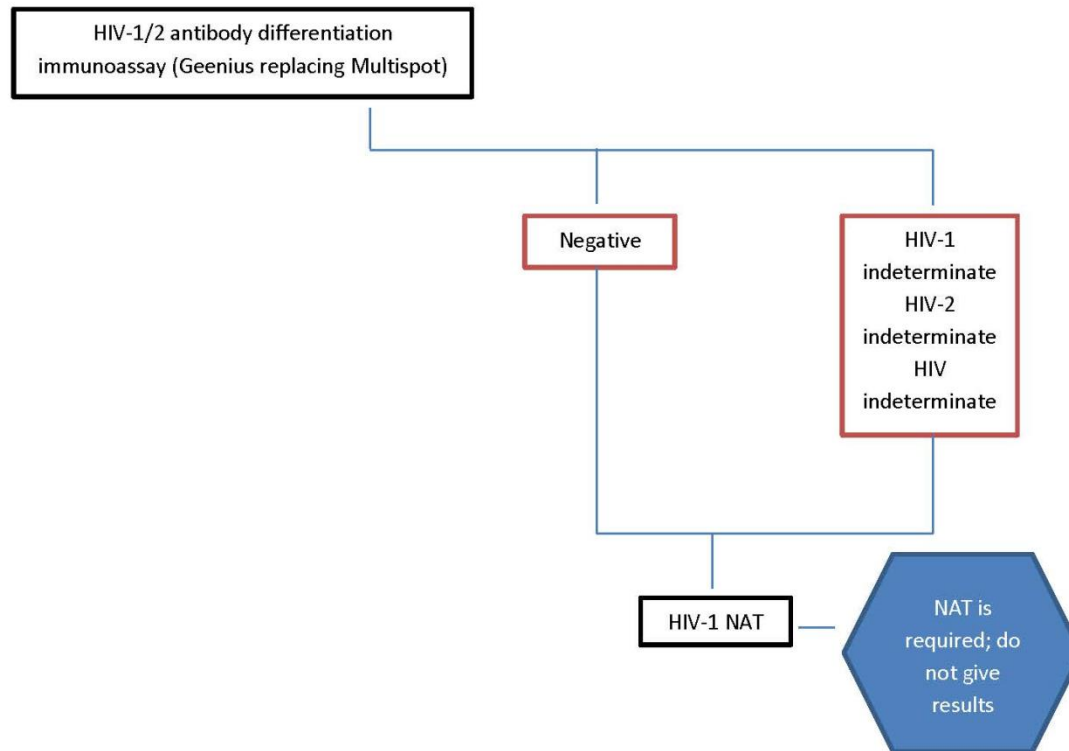
Here's Where It Starts...



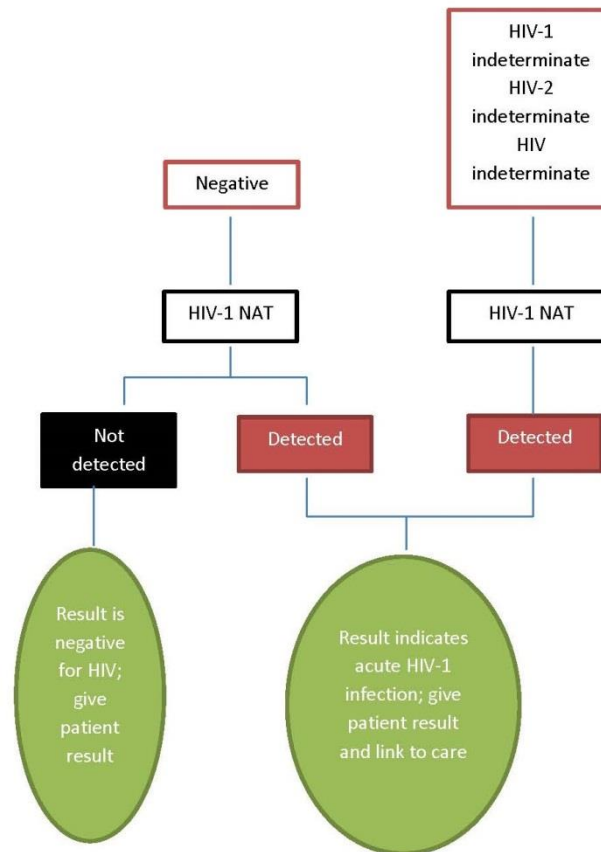
...And Here's Where Antibodies Are Detected...



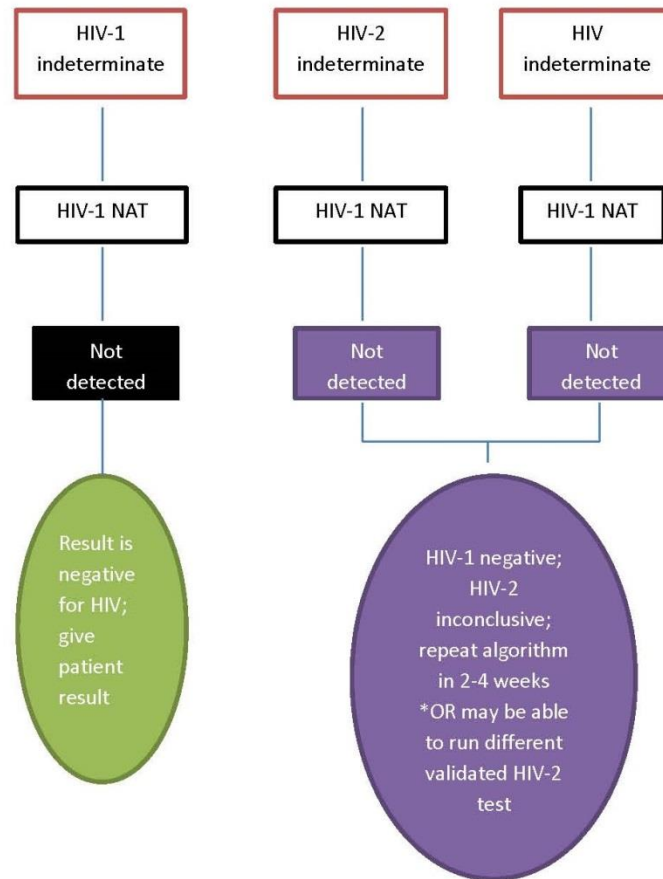
...And Here's Where More Testing Is Needed...



Possible NAT Results...



...And What About Indeterminates?



Goal of the HIV Laboratory Testing Algorithm

- According to the Association of Public Health Laboratories, the goal is “to maximize the identification of new, previously undiagnosed HIV infections”.

Lab Result: Negative

Sample Information

SAMPLE ID: 1674406
CLINICIAN: PARPART,FAYE
CLINICIAN PHONE:
SOURCE OF SPECIMEN: BLOOD VENOUS
SOURCE TYPE:
PROGRAM: CLINIC
SUBPROGRAM: VD

DATE COLLECTED: 01/20/2017
DATE SUBMITTED: 01/20/2017
DATE RECEIVED: 01/21/2017
SPECIMEN SITE:
SITE TYPE: STD
VDH OUTBREAK #:
MEDICAL RECORD #:
GRANT CODE :

TEST RESULTS

HIV Combo Ag/Ab EIA

| Test | Results |
|------------------------|--|
| HIV 1+2 Ab+HIV1 p24 Ag | Non-reactive - Negative for the presence of detectable HIV-1 p24 antigen, HIV-1 antibodies (Groups M and O) and HIV-2 antibodies by EIA. |

Information Provided by Submitter

| Questions | Responses |
|----------------------------------|--------------|
| Has a rapid test been performed? | NO |
| Who is the attending Clinician? | PARPART,FAYE |

Counseling messages

- Negative screening test:
 - HIV not detected, return for re-testing in one month if there is concern of having been exposed to the infection within the past month
- Positive **screening** test:
 - **We will not know** because we do not get these back without the Geenius
 - There would be nothing we could tell the patient at this point because we do not have a completed algorithm

Lab Result: Positive

TESTING PERFORMED BY
Division of Consolidated
Laboratory Services (DCLS)
800 N. 8th Street
Richmond, VA
804-648-4480

RESULTS ELECTRONICALLY
RECEIVED BY
Virginia Department of Health (VDH)
109 Governor Street
Richmond, VA 23219
804-884-7200 x 2

FINAL REPORT

Report Date: 04/24/2017
DCLS LIMS #: R170419801

Printed By

RICHMOND CITY HEALTH DISTRICT
STE 442-A 400 E CARY ST
RICHMOND, VA 23219
HEALTH DEPT SITE CODE: 760E

Submitted By

RICHMOND CITY HEALTH DISTRICT
STE 442-A 400 E CARY ST
RICHMOND, VA 23219
PHONE: 804-206-3500

Patient Information

[REDACTED]

DOB: [REDACTED]
SEX: [REDACTED]
RACE: [REDACTED]

PATIENT ID: [REDACTED]

Sample Information

SAMPLE ID: 1721802
CLINICIAN: PARPART, FAY
CLINICIAN PHONE:
SOURCE OF SPECIMEN: BLOOD VENOUS
SOURCE TYPE:
PROGRAM: CLINIC
SUBPROGRAM: VD

DATE COLLECTED: 04/17/2017
DATE SUBMITTED: 04/17/2017
DATE RECEIVED: 04/18/2017
SPECIMEN SITE:
SITE TYPE: STD
VDH OUTBREAK #:
MEDICAL RECORD #:
GRANT CODE:

TEST RESULTS

HIV Combo Ag/Ab EIA

Test

Genius HIV-1/HIV-2

HIV 1+2 Ab+HIV1 p24 Ag

Results

HIV - 1 Positive - The test result is POSITIVE for HIV-1 by the Genius HIV1/2 Supplemental Assay using an immunodotmembrane method. A positive assay result confirms the presence of antibodies to HIV-1.

Reactive - Repeatedly reactive for the presence of HIV-1 p24 antigen, HIV-1 antibodies (Groups M and O) and/or HIV-2 antibodies by EIA.

CONFIDENTIALITY NOTICE: This report contains PRIVILEGED and CONFIDENTIAL INFORMATION. It is intended solely for the exclusive use of VDH and the intended recipient(s). If you received this report in error, please notify VDH immediately at (804) 884-7200 x 2.

Printed on: 04/25/2017

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Division of Consolidated
Laboratory Services (DCLS)
800 N. 5th Street
Richmond, VA
804-648-4480

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STE 442-A 400 E CARY ST
RICHMOND, VA 23219

HEALTH DEPT SITE CODE: 760E

Submitted By

RICHMOND CITY HEALTH DISTRICT
STE 442-A 400 E CARY ST
RICHMOND, VA 23219

PHONE: 804-206-3500

Patient Information

[REDACTED]

DOB: [REDACTED] AGE: [REDACTED]
SEX: [REDACTED]
RACE: [REDACTED]

PATIENT ID: [REDACTED]

POST TEST COUNSELING: [REDACTED] COUNSELOR #: [REDACTED] DATE: [REDACTED]
[REDACTED]

Sample Information

SAMPLE ID: 1721652
CLINICIAN: PARPART, FAY
CLINICIAN PHONE:
SOURCE OF SPECIMEN: BLOOD VENOUS
SOURCE TYPE:
PROGRAM: CLINIC
SUBPROGRAM: VD

DATE COLLECTED: 04/17/2017
DATE SUBMITTED: 04/17/2017
DATE RECEIVED: 04/18/2017
SPECIMEN SITE:
SITE TYPE: STD
VDH OUTBREAK #:
MEDICAL RECORD #:
GRANT CODE :

TEST RESULTS

HIV Combo Ag/Ab EIA

| Test | Results |
|------------------------|---|
| Genius HIV-1/HIV-2 | HIV - 1 Positive - The test result is POSITIVE for HIV-1 by the Genius HIV1/2 Supplemental Assay using an immunochromatographic method. A positive assay result confirms the presence of antibodies to HIV-1. |
| HIV 1+2 Ab+HIV1 p24 Ag | Reactive - Repeatedly reactive for the presence of HIV-1 p24 antigen, HIV-1 antibodies (Groups M and O) and/or HIV-2 antibodies by EIA. |

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Counseling messages—Positive for HIV 1 or HIV 2

- Your HIV test is positive, which means you have HIV infection. It's important that you see a doctor for a medical evaluation. I can help you set that up.
- Your HIV test is positive for HIV-2, which is more common in certain parts of the world (such as West Africa). It's important that you see a doctor, and I can help you make an appointment. It's also very important that you tell the doctor that your test is positive for HIV-2, as that will affect decisions regarding your medical care.

New Results

- HIV-2 reactive with HIV-1 cross reactivity: This is considered HIV-2 positive
 - Link patient to care
- HIV positive-untypable (undifferentiated): Positive for HIV-1 and HIV-2 antibodies; evidence of HIV-1 and/or HIV-2 infection is present
 - Link patient to care; provider may consider additional testing to verify or rule out dual infection

Lab Result: Algorithm Incomplete!!

TEST RESULTS

HIV Combo Ag/Ab EIA

HIV 1+2 Ab+HIV1 p24 Ag

Reactive - Repeatedly reactive for the presence of HIV-1 p24 antigen, HIV-1 antibodies (Groups M and O) and/or HIV-2 antibodies by EIA.

Test

Results

Genius HIV-1/HIV-2

HIV Negative - No serological evidence of HIV-1 and HIV-2 by the Genius HIV1/2 Supplemental Assay using an immunochromatographic method. A negative or indeterminate result does not preclude the possibility of exposure to HIV or infection with HIV. The sample will be sent for Nucleic Acid Amplification testing (NAAT).

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Printed on: 04/25/2017

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Lab Result: Algorithm Incomplete!!

TESTING PERFORMED BY
Division of Consolidated
Laboratory Services (DCLS)
600 N. 6th Street
Richmond, VA
804-648-4480

RESULTS ELECTRONICALLY
RECEIVED BY
Virginia Department of Health (VDH)
108 Governor Street
Richmond, VA 23219
804-864-7200 x 2

FINAL REPORT

Report Date: 04/24/2017
DCLS LIMS #: R170419940

Printed By

RICHMOND CITY HEALTH DISTRICT
STE 442-A 400 E CARY ST
RICHMOND, VA 23219
HEALTH DEPT SITE CODE: 7806

Submitted By

RICHMOND CITY HEALTH DISTRICT
STE 442-A 400 E CARY ST
RICHMOND, VA 23219
PHONE: 804-205-9500

Patient Information

PATIENT ID: [REDACTED]

DOB: [REDACTED] AGE: [REDACTED]
SEX: [REDACTED]
RACE: [REDACTED]
ETHNICITY: [REDACTED]
MARITAL STATUS: [REDACTED]

Sample Information

SAMPLE ID: 1721604
CLINICIAN: PAPPART, FAY
CLINICIAN PHONE:
SOURCE OF SPECIMEN: BLOOD VENOUS
SOURCE TYPE:
PROGRAM: CLINIC
SUBPROGRAM: VD

DATE COLLECTED: 04/17/2017
DATE SUBMITTED: 04/17/2017
DATE RECEIVED: 04/18/2017
SPECIMEN SITE:
SITE TYPE: STD
VDH OUTBREAK #:
MEDICAL RECORD #:
GRANT CODE:

TEST RESULTS

HIV Combo Ag/Ab EIA

HIV 1+2 Ab+HIV1 p24 Ag

Reactive - Repeatedly reactive for the presence of HIV-1 p24 antigen, HIV-1 antibodies (Groups M and O) and/or HIV-2 antibodies by EIA.

Test

Genieus HIV-1/HIV-2

Results

HIV Negative - No serological evidence of HIV-1 and HIV-2 by the Genieus HIV1/2 Supplemental Assay using an immunochromatographic method. A negative or indeterminate result does not preclude the possibility of exposure to HIV or infection with HIV. The sample will be sent for Nucleic Acid Amplification testing (NAAT).

Counseling messages

- Your HIV results are not back yet; we need to schedule another time for you to return. In the meantime, it's important for you to take precautions to protect yourself and others – use condoms for vaginal, oral and anal sex, and if you inject drugs, don't share needles or works with anyone.
- This is one reason it is important to tell the patients that it takes 2 weeks to get results

Lab slip: Negative NAT

Date received: 12/28/2016

FINAL LABORATORY REPORT

Report Date

Bloodborne Viruses Laboratory

Phone: (518) 474-2163 Fax: (518) 473-0008

Testing performed at CLIA# 33D2005937

Specimen Id: RIL1636300010

Specimen Type: Serum

Qualitative HIV-1 RNA:

HIV-1 RNA Not Detected

12/30/2016

END OF REPORT

Counseling messages

- HIV not detected, return for re-testing in one month if there is concern of having been exposed to the infection within the past month

Lab slip: Positive NAT

FINAL LABORATORY REPORT

Bloodborne Viruses Laboratory

Specimen Id: RIL1607500012

120 New Scotland Avenue
Albany, New York 12208
Phone: (518) 474-2163 Fax: (518) 473-0008

Specimen Type: Serum

Qualitative HIV-1 RNA (TMA) :

HIV-1 RNA Detected

The APTIMA HIV-1 Qualitative Assay is an FDA-approved nucleic acid assay using transcription-mediated amplification (TMA) for the detection of HIV-1 in human plasma and serum.

END OF REPORT

Counseling messages

- Your HIV test is positive, and shows you likely got HIV recently. Based on your history and this test result, it is important that you see a doctor very soon. It is important that you know that HIV is very easily transmitted at this stage. I'd like to assist you with a referral to a doctor.
- This indicates likely acute infection. Link patient to care or to a patient navigator
 - An expedited visit should be scheduled

What If Geenius Is Indeterminate and NAT Is Not Detected?

- If Geenius is indeterminate for HIV-1 and NAT is not detected, the final result is negative
- If Geenius is indeterminate for HIV or indeterminate for HIV-2, and NAT is not detected, the result is HIV-1 negative, HIV-2 inconclusive
 - Redraw in 2-4 weeks and repeat algorithm
 - *OR may be able to run different validated HIV-2 test--stay tuned for more information

4th Generation HIV Algorithm

Benefits

- Detects IgM & IgG Antibodies; p24 Antigen
- 4th gen IA detects HIV-1 approximately 1 week earlier than 3rd gen assay
- Addition of HIV-1 RNA NAT helps detect false positive results & increases sensitivity to detect acute HIV infection
- Geenius assay differentiates between HIV-1 and HIV-2

4th Generation HIV Algorithm

Benefits (con't.)

- Once linked to care & on ARV therapy, can result in better health outcomes for the patient
- Reduces likelihood of spreading infection
 - Individuals are less likely to spread infection if they are aware they are infected
 - Once on ARV therapy, greater chance of achieving undetectable viral load

Why Does Understanding the WHOLE Algorithm Matter?

- If **NOT** understood...
 - Could lead to misinterpretation of results by health care providers
 - Unnecessary additional testing may be ordered
 - Could lead to delay in accessing care

Questions?

- Got it?
- ...or starting to get it?
- GOOD!!!

- Caroline Campbell
 - 804-864-7978 or Caroline.Campbell@vdh.virginia.gov

- Kendra Weindling
 - 804-482-8007 or Kendra.Weindling@vdh.virginia.gov